In the name of God

Exosome in Cancer Disease

Affiliation: Qazvin University of medical sciences

school of paramedical

Presented by: Farhangi . A

Under Supervision : Dr. Ahmadpour

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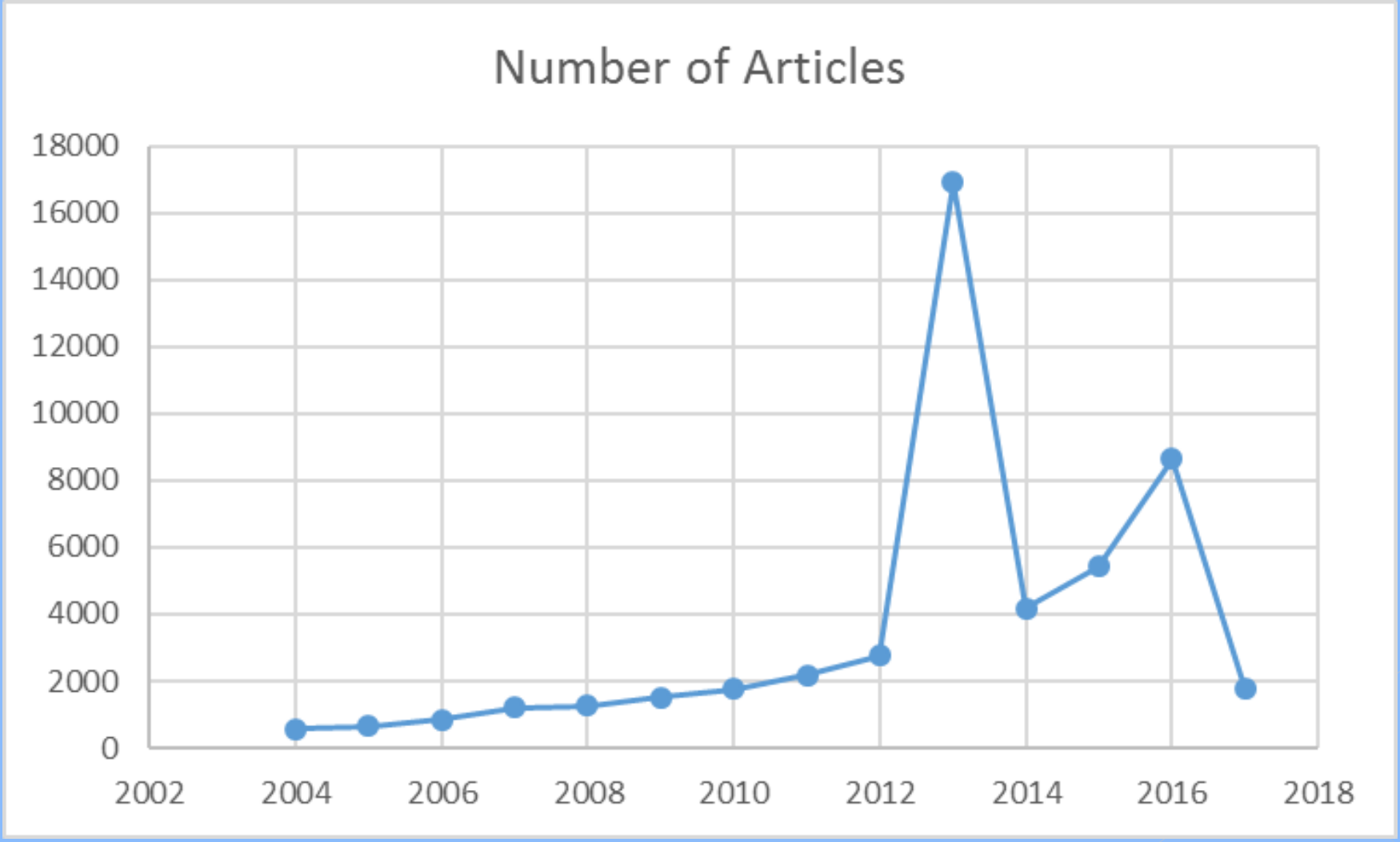
Research trend



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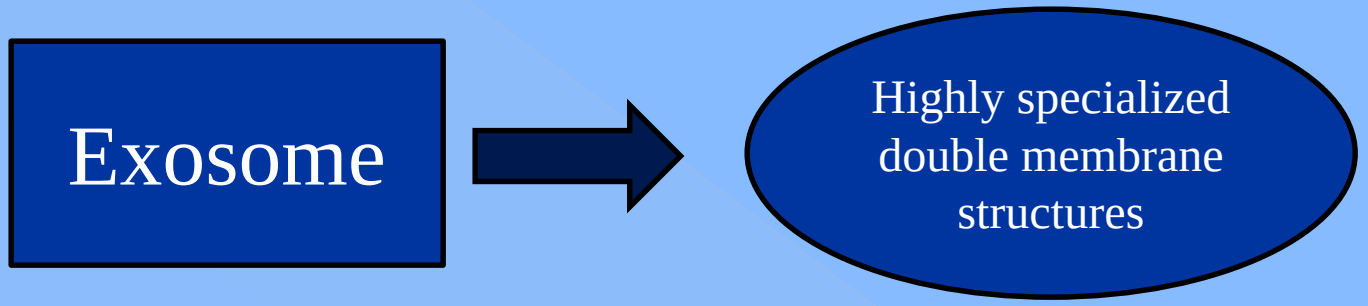
Introduction

Introduction

- Definition



- Exosomes are involved in intracellular communication

Introduction

- Tumor cells have various mechanisms against immune system

  Such as secretion of exosomes

TEX: Tumor-derived Exosomes

Exosomes can be carriers of various proteins, lipids, miRNAs and m RNAs

Introduction

- The role of exosomes

influence in metastasis and cancer progression

involve in multidrug resistance mechanisms

Enables cancer cells to evade from recognition by host immune cells

Introduction

- The role of exosomes

role as communication vehicles between cells

Responsible for the formation of vessels

Prognosis of cancer

Introduction

- **Exosome: source of information concerning prognosis**

Patient condition and the effectiveness of applied treatment

Introduction

Source of exosome

Exosome can be obtained from:

- **Amniotic fluids**
- **ascites**
- **nasal lavage fluid**
- **Serum**
- **Plasma**
- **Milk**
- **Urine**
- **CSF**
- **Cell culture medium**

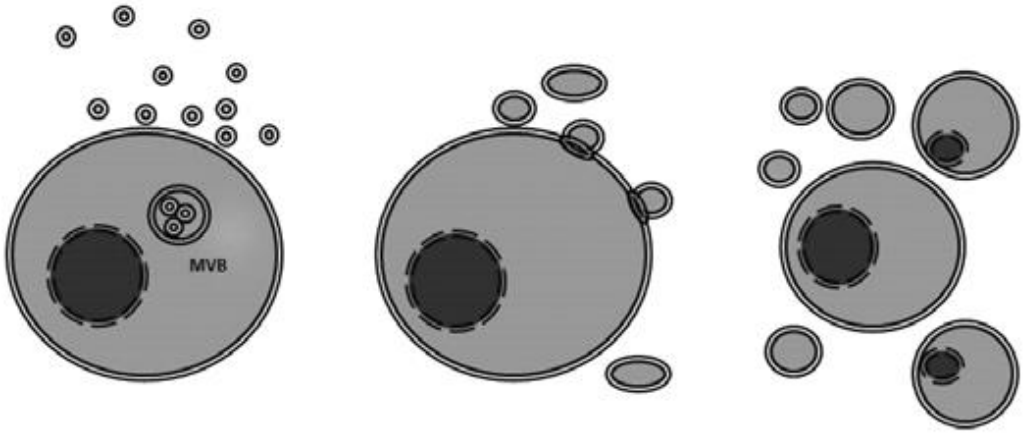
Introduction

- **Factors affecting the content and amount of vesicles:**
  - **Physical factors: ionizing radiation, heat**
  - **Chemical factors: low pH level, increased concentration of calcium, oxidative stress, hypoxia**

The Origin of Exosomes

The origin of exosomes

- **The type and biogenesis of different extracellular vesicles**



	Exosomes	Microvesicles	Apoptotic bodies
Size of vesicles / Shape	30- 100 nm, regular	100- 1000 nm, irregular	50- 5000 nm, irregular
Markers	LAMP-1, tetraspanins, Alix, MHC -I, -II, HSP70, TSG100	Selectins, integrins, tissues factor and cell-specific markers	Histones, organelles
Origin	Endosomal compartments of cells	Cell surface plasma membrane	Cells which undergo apoptosis

The role of exosomes in tumor progression and metastasis (Review)

The origin of exosomes

- **ILV:**

  Intra Luminal Vesicles

- **MVB:**

  Multi Vesicular Body

The origin of exosomes

- **MVB: an intracellular compartment containing multiple vesicles with the plasma membrane.**

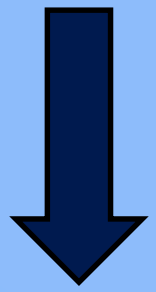
MVBs are assembled from vesicles sorted from the trans- Golgi network or from internalized membrane

The origin of exosomes

- MVB pathways variants:

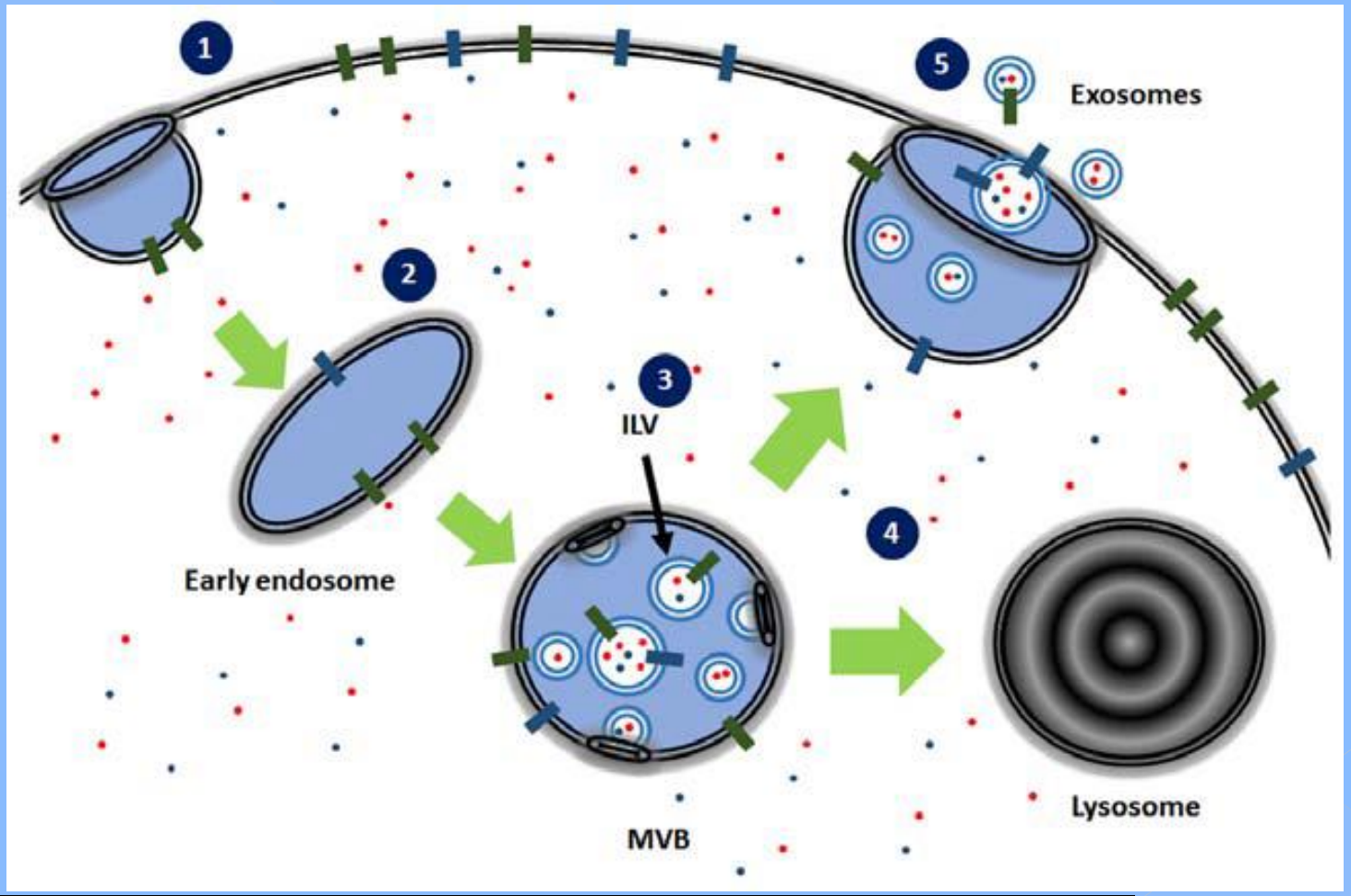
MVB can fuse with lysosomes for protein degradation

Or release their ILV by fusing with the plasma membrane.



The release vesicles are termed exosomes

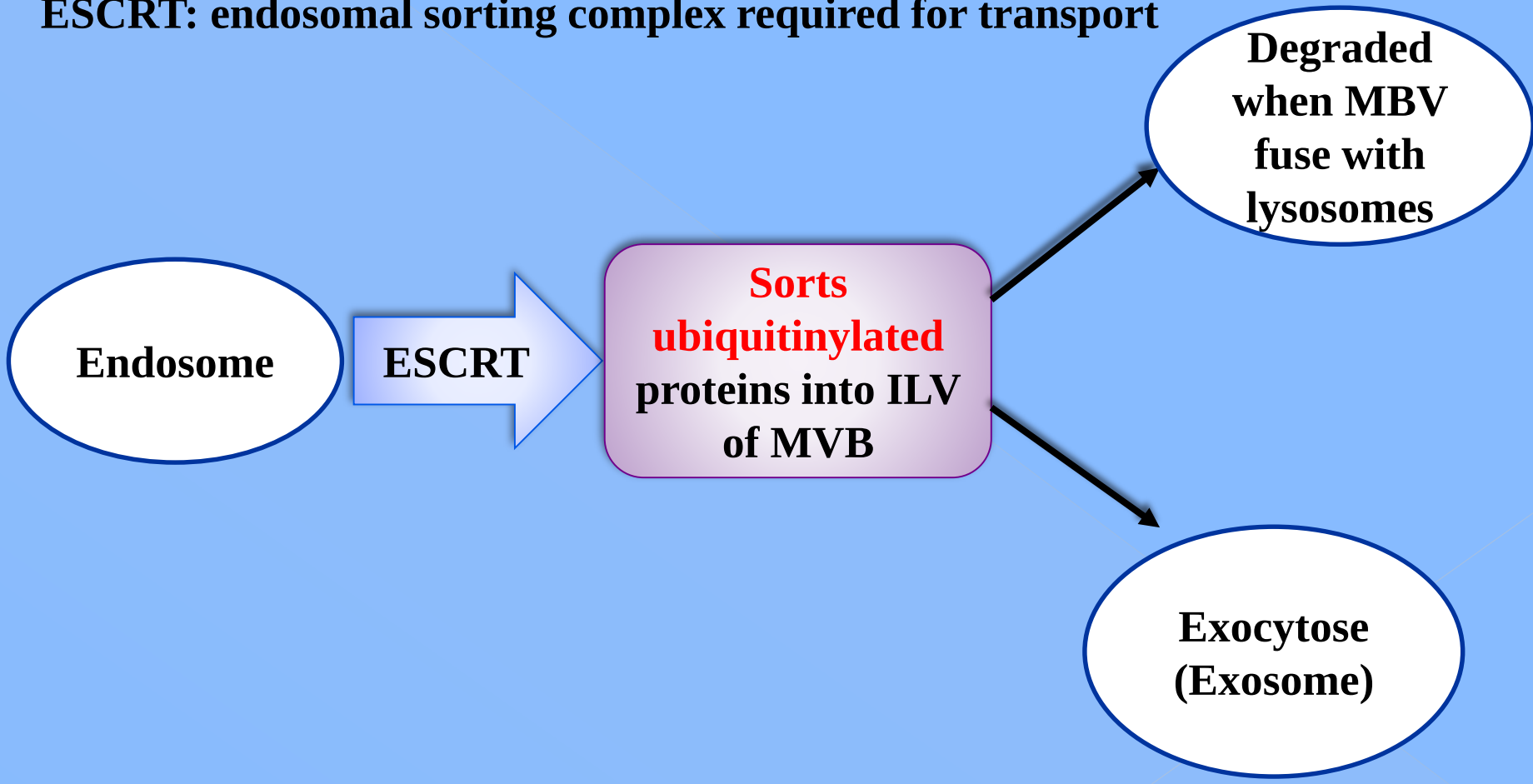
The origin of exosomes



The role of exosomes in tumor progression and metastasis (Review)

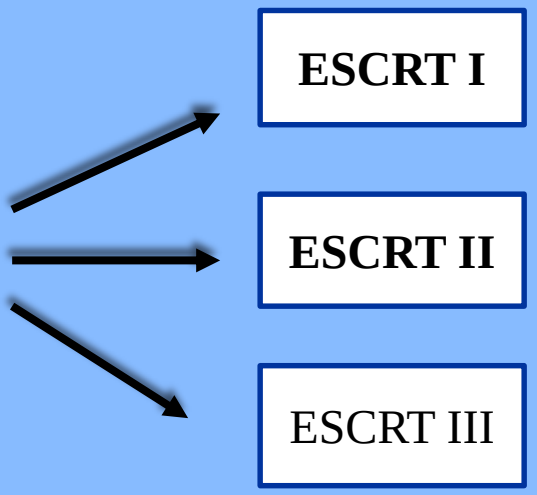
The origin of exosomes

ESCRT: endosomal sorting complex required for transport



The origin of exosomes

- ESCRT complex
- The sub complex of ESCRT complex

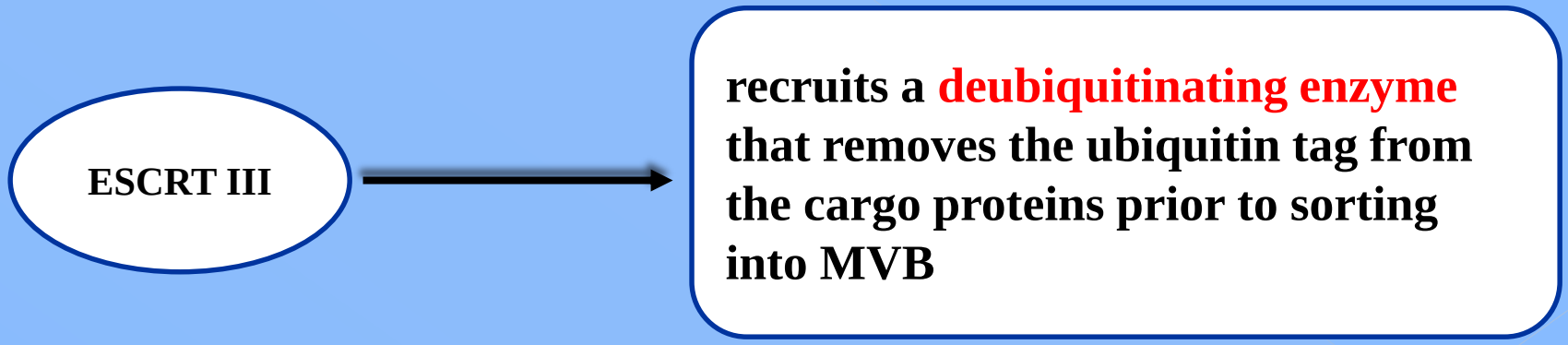


Tsg 101 (tumor susceptibility gene) in the ESCRT complex I

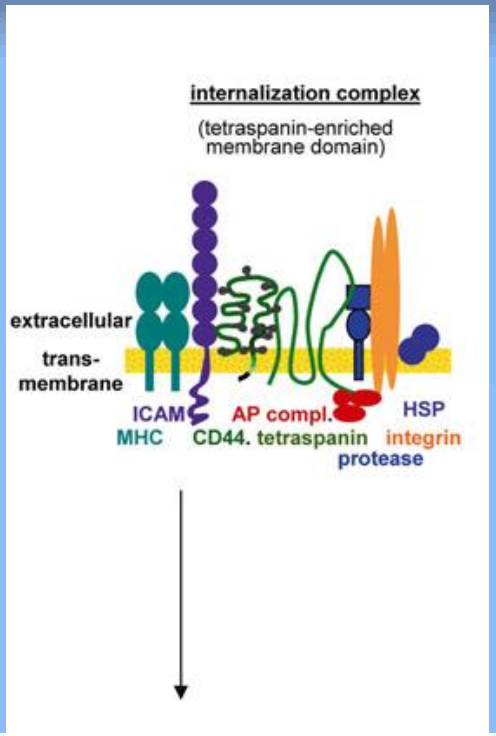
binds ubiquitinated proteins

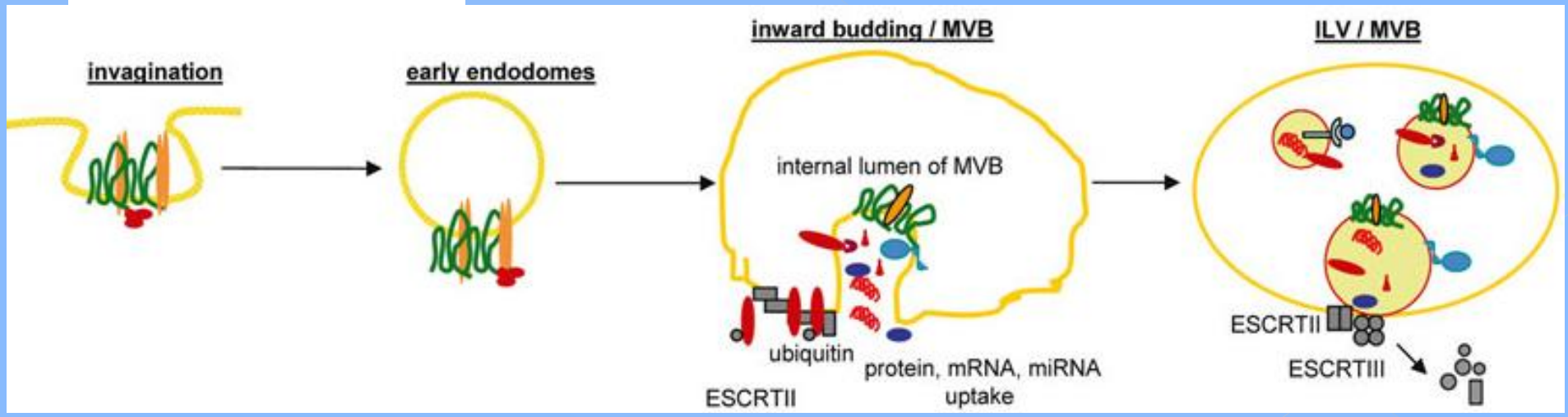
Recruits ESCRT II

The origin of exosomes



The origin of exosomes





The origin of exosomes

APC (antigen presenting cells)

release exosomes derived from the Major histocompatibility (MHC) class II compartment, a subset of MVB.

Exosome containing RNA or miRNA

induce exogenous gene expression and mediate gene silencing

Exosome Characterization

Exosome characterization

- **Exosome constituents: Exosomes are composed of a lipid bilayer enriched in cholesterol, sphingomyelin,GM3, and phosphatidylserine.**

Constitutive membrane components are tetraspanins, adhesion molecules, proteases, and trans membrane receptors

- **Exosomes are 50–100 nm in size and, according to the lipid composition, have a density of 1.14–1.17 g/l**

Exosome Isolation

Exosome isolation

- **Exosomes being released in the extracellular space are purified from cell culture supernatants and biological fluids**

- **Commonly used isolation methods:**

ultracentrifugation at 100,000 × g for 1–2 h to pellet the Exosomes Size chromatography preferably HPLC

Exosome and Cancer

Role of exosome in cancer

- **Role of TEX in immune suppression**

they carry immunosuppressive molecules and factors known to interfere with immune cell functions

TEX also deliver genomic DNA, mRNA, and microRNAs to immune cells, thereby reprogramming functions of responder cells to promote tumor progression. TEX carrying tumor-associated antigens can interfere with antitumor immunotherapies.

Mechanisms of TEX – mediated immune suppression

- **TEX carry inhibitory ligands that bind to cognate receptors on immune cells, inducing negative signaling .**

- **The two key receptors on immune cells, the T cell receptor (TCR) and the IL-2 receptor (IL-2R), are negatively regulated by TEX.**

Mechanisms of TEX – mediated immune suppression

- **The effect on T lymphocyte**

inhibition of CD3ζ chain expression and reduced levels of mRNA coding for the CD3ζ chain

(CD 3 ⟶ T lymphocytes phenotypic marker)

Mechanisms of TEX – mediated immune suppression

- **The effect on T lymphocyte**

TEX reduce JAK kinase expression in activated T cells .The integrity of the JAK pathway is critical for the functions of cytokines sharing the γ-chain of the IL-2R (IL-2, IL-7, IL-15);

thus, suppression of IL-2R γ-chain phosphorylation levels leads to the failure of T cells to produce these cytokines and to proliferate

Mechanisms of TEX – mediated immune suppression

- **The effect on T $CD8^+$**

Experiments showed that TEX inhibited the proliferation of human CD8+ T cells

Mechanisms of TEX – mediated immune suppression

- **TEX induce apoptosis of activated CD8+ T effector**

Nearly all CD8+ T lymphocytes in the circulation of cancer patients express surface CD95 (Fas receptor), and many express programmed death 1 (PD-1)

Therefore, they are susceptible to apoptosis by TEX carrying the membrane form of FasL or programmed death ligand 1 (PD-L1), respectively.

Mechanisms of TEX – mediated immune suppression

- **The effect on NK cell**

TEX suppress NK cell activity. The frequency and activity of NK cells are often depressed in cancer patients compared with age-matched, healthy individuals.

Mechanisms of TEX – mediated immune suppression

- **The effect on NK cell**

Additionally, expression levels of the NK cell–activating receptors NKp30, NKp46, NKG2C, and NKG2D are low in cancer patients . TEX downregulate expression of NKG2D and reduce NK cell cytotoxicity

Mechanisms of TEX – mediated immune suppression

- **The effect on monocyte**

TEX interfere with monocyte differentiation. Co incubation of peripheral blood monocytes (PBMCs) with TEX promoted their differentiation into TGF-β–expressing DCs,

which also secreted PGE2 and interfered with cytotoxic T lymphocyte (CTL) generation.

Mechanisms of TEX – mediated immune suppression

- **The effect on monocyte**

DCs generated in the presence of TEX expressed low levels of costimulatory molecules and induced dose-dependent inhibition of T cell proliferation.

Mechanisms of TEX – mediated immune suppression

- **The effect on myeloid precursor**

TEX skew the differentiation of myeloid precursor cells into **MDSCs**

(myeloid – derived suppressor cells)

MDSC accumulation has a two-fold effect on the immune response: first, with the paucity or absence of DCs, antigen processing and presentation are negatively affected,

Mechanisms of TEX – mediated immune suppression

- **The effect on myeloid precursor**

and, second, the newly minted MDSCs produce numerous immunosuppressive inhibitory factors, including NO and ROS, which cause nitration of TCRs or T cell apoptosis

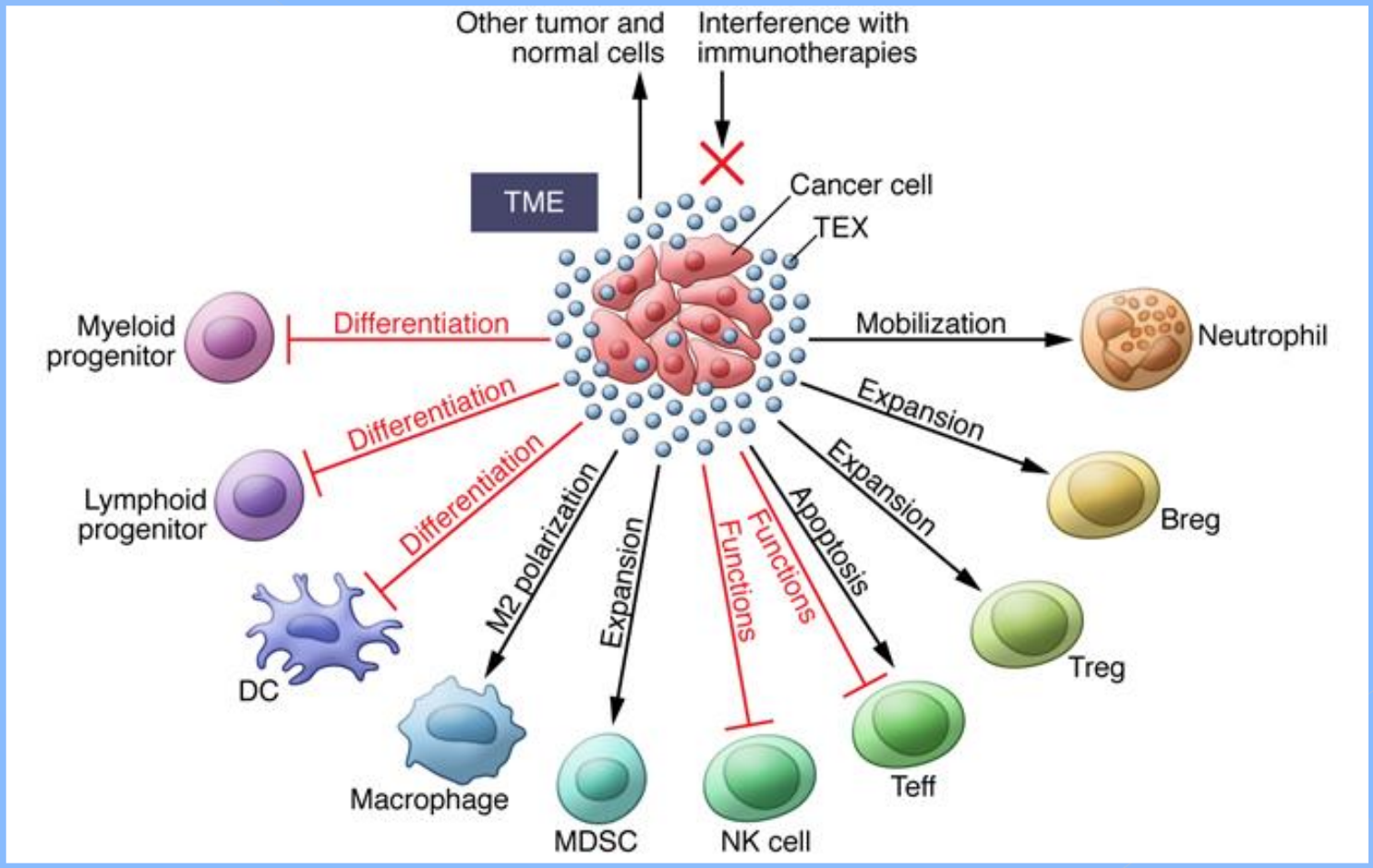
Mechanisms of TEX – mediated immune suppression

- **The effect on T regulator**

TEX drive differentiation and expansion of Tregs. The frequency of circulating $CD4^+CD25^+FOXP3^+$ Tregs is often elevated in patients with cancer

TEX induced the conversion of human conventional $CD4^+CD25^-$ T cells to $CD4^+CD25^+FOXP3^+$Tregs

Mechanisms of TEX – mediated immune suppression



Exosomes and tumor-mediated immune suppression (Review)

TEX interfere with cancer immunotherapies

- **As TEX are known to carry TAAs (Tumor Associated Antigens), they can efficiently bind and sequester tumor-reactive Abs and dramatically reduce binding of these Abs to tumor cells.**

Exosome and Cancer

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The Journal of Clinical Investigation

Exosomes and tumor-mediated immune suppression

Theresa L. Whiteside

Departments of Pathology, Immunology, and Otolaryngology, University of Pittsburgh School of Medicine and University of Pittsburgh Cancer Institute, Pittsburgh, Pennsylvania, USA.

Tumor-derived exosomes (TEX) are harbingers of tumor-induced immune suppression: they carry immunosuppressive molecules and factors known to interfere with immune cell functions. By delivering suppressive cargos consisting of proteins similar to those in parent tumor cells to immune cells, TEX directly or indirectly influence the development, maturation, and antitumor activities of immune cells. TEX also deliver genomic DNA, mRNA, and microRNAs to immune cells, thereby reprogramming functions of responder cells to promote tumor progression. TEX carrying tumor-associated antigens can interfere with antitumor immunotherapies. TEX also have the potential to serve as noninvasive biomarkers of tumor progression. In the tumor microenvironment, TEX may be involved in operating numerous signaling pathways responsible for the downregulation of antitumor immunity.

Exosome as a diagnostic tool

Exosome as a diagnostic tool

- **They are released by the majority of the cells and contain detailed molecular information of the tumor cells**

- **Exosomes can be isolated from easy accessible body fluids, and most importantly, they can provide several biomarkers, with different levels of specificity**

Exosome as a diagnostic tool

- **Recent clinical evidence shows that the levels of Exosomes released into body fluids may themselves represent a predictive/diagnostic of tumors**

Exosome as a diagnostic tool

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Exosome levels in human body fluids: A tumor marker by themselves?

Francesco Cappello [a,b], Mariantonia Logozzi [c], Claudia Campanella [a,b], Celeste Caruso Bavisotto [a,b], Antonio Marcilla [d,e], Francesca Properzi [c], Stefano Fais [c,*]

[a] Department of Experimental Biomedicine and Clinical Neuroscience, University of Palermo, Palermo, Italy
[b] Euro-Mediterranean Institute of Science and Technology, Palermo, Italy
[c] Department of Therapeutic Research and Medicines Evaluation, National Institute of Health, Rome, Italy
[d] Àrea de Parasitologia, Departament de Biologia Cellular i Parasitologia, Universitat de València, Burjassot, Valencia, Spain
[e] Joint Research Unit on Endocrinology, Nutrition and Clinical Dietetics, Health Research Institute La Fe, Universitat de Valencia, Valencia, Spain

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ABSTRACT

Despite considerable research efforts, the finding of reliable tumor biomarkers remains challenging and unresolved. In recent years a novel diagnostic biomedical tool with high potential has been identified in extracellular nanovesicles or exosomes. They are released by the majority of the cells and contain detailed molecular information on the cell of origin including tumor hallmarks. Exosomes can be isolated from easy accessible body fluids, and most importantly, they can provide several biomarkers, with different levels of specificity. Recent clinical evidence shows that the levels of exosomes released into body fluids may themselves represent a predictive/diagnostic of tumors, discriminating cancer patients from healthy subjects. The aim of this review is to highlight these latest challenging findings to provide novel and groundbreaking ideas for successful tumor early diagnosis and follow-up.

exosome as therapeutics

exosome as therapeutics

- **They are small and flexible, which allows them to cross biological membranes.**

- **By a lipid bilayer they protect their cargo from degradation.**

exosome as therapeutics

- **The capacity of exosomes to serve as bio vesicles for nucleic acids, proteins, and lipids, and their role in intercellular communication, make them a versatile platform for drug delivery**

exosome as therapeutics

- **Exosomes are intrinsically bioactive; thus, they can be isolated from cells for downstream use without further modification**

- **modification of exosomes allowing researchers to specifically customize or tailor exosomes to particular applications**

exosome as therapeutics

- **Exosome as therapeutic in Alzheimer's disease**

Alvarez-Erviti and colleagues were the first to enrich exosomes with siRNAs and genetically equip them with neuron-specific rabies virus glycoprotein (RVG) peptides to target the brain

They were able to silence 60% of beta-secretase 1 (BACE-1) expression, a key player in the progression of Alzheimer's disease

exosome as therapeutics

- **Since then, a variety of studies have reported the use of exosomes for the treatment in various diseases including in :**

oncology, neurology, ischemic diseases, liver disease, therapeutic vaccination, and immune disorders

Conclusion

Conclusion

- **TEX are rapidly emerging as a critical component of a tumor orchestrated information system that is designed to facilitate tumor immune escape and promote tumor growth**

- **Unique exosomal cargo contents can be used in the future as potential predictive biomarkers, which enable the observation of patients before and during treatment**

Conclusion

- **However, the specific separation of only one extracellular membrane vesicle subpopulation is problematic**

due to the wide size range, and characterization of their content remains a challenge for researchers.

References

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- **The role of exosomes in tumor progression and metastasis (Review)/Wiktoria M. Suchorska1,3 and Michal S. Lach1,2 / 2016**